USE OF LICORICE EXTRACT IN COUNTERACTING AFLATOXICOSIS IN BROILERS

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ABSTRACT

This study was conducted at the Animal Production Farm/State Board for Agricultural Research over the period from 21/3/2002 to 27/4/2002 .A total of 900 Fawbro broilers, three weeks - old were used to investigate the probable role of licorice extract in suppressing the deterimental effects of aflatoxicosis on productive performance of broilers.Chicks were randomly allocated to 6 treatments of 3 replicates.Birds in the first treatment (T1) were fed a basal diet and used as control group. Birds in T2 treatment fed a diet contaminated with aflatoxin, while birds in T3 treatment were fed a diet contaminated with aflatoxin and treated with mold killer. However, birds in T4, T5 and T6 treatments were fed a diet contaminated with aflatoxin and supplemented with licorice extract at the levels of 150, 300 and 450mg/kg of diet, respectively.

Inclusion of the aflatoxin in the diet resulted in a significant ($p<0.05$) decrease in body weight, weight gain, feed consumption, feed conversion efficiency, Productive Index, Economic Figure, dressing percentage with or without viscera; an enlarged liver, spleen, heart, and gizzard and increased in abdominal fat. When mold killer (T3) or licorice extract (T4, T5 and T6) were incorporated into the diet containing aflatoxin, they significantly ($p<0.05$) improved all these traits. However, licorice treatments surpasses T3 as regards live body weight, weight gain, Productive Index and Economic Figure. Furthermore, there was a trend that T6 recorded the best results in relation to productive characteristics included in this study in comparison with T3, T4 and T5 treatments.

It was concluded from this study that supplementation of licorice extract particularly at a level of 450 mg/kg to the diet contaminated with aflatoxin can depress the adverse effects of aflatoxicosis on productive performance of broiler chickens.

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استخدام مستخلص عرق السوس لتقليل آثار التسمم بسموم الافلاتوكسين في فروج اللحم

حازم جبار الدراجي
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المستخلص

اجريت هذه الدراسة في حقل الانتاج الحيواني التابع للهيأة العامة للبحوث الزراعية/وزارة الزراعة للمدة من 2002/3/21 لغاية 2002/4/27 وأستخدم فيها 900 فرخ فروج لحم فاوبرو بعمر ثلاثة اسابيع لبحث الدور المحتمل لمستخلص عرق السوس في تقليل الآثار الضارة الناجمة عن تسمم العلف بسموم الافلاتوكسين. تم توزيع الافراخ عشوائياً على ثلاثة معاملات يتكون كل منها من ثلاثة مكررات وبواقع 150 فرخا لكل معاملة . وتم تغذية الطيور في المعاملة الاولى (T1) على عليقة السيطرة وعدت كمجموعة مقارنة . وغذيت الطيور في المعاملة T2 على العليقة الملوثة بسموم الافلاتوكسين ، في حين ان طيور المعاملة T3 غذيت على علية ملوثة بسموم الافلاتوكسين ومعاملة بماده قاتلة للفطريات Mold killer . من ناحية ثانية ، فأن الطيور في المعاملات T4 و T5 و T6 غذيت على عليقة ملوثة بسموم الافلاتوكسين ومضاف اليها مستخلص عرق السوس بمستويات 150 و 300 و 450 ملغم/كغم علف على التوالي.

اشارت النتائج الى ان تلوث العلف بسموم الافلاتوكسين ادى الى انخفاض معنوي (أ<0.05) في الوزن الحي للطير والزيادة الوزنية واستهلاك العلف وكفاءة التحويل الغذائي والدليل الانتاجي والمؤشر الاقتصادي ونسبة التصافي مع او بدون الاحشاء الداخلية والى زيادة معنوية (أ<0.05) في الوزن النسبي للكبد والطحال والقلب والقانصة ودهن البطن. من ناحية ثانية ، عندما تمت اضافة المادة القاتلة للعفن (T3) او عرق السوس (T4 و T5 و T6) للعلف الملوث بسموم الافلاتوكسين فأنها حسنت معنوياً (أ<0.05) جميع الصفات المذكورة بالمقارنة مع المعاملة T2. كما ان معاملات عرق السوس قد تفوقت على المعاملة T3 فيما يتعلق بوزن الجسم الحي والزيادة الوزنية والدليل الانتاجي والمؤشر الاقتصادي . فضلاً على ذلك ، كان هناك اتجاه واضح بأن المعاملة T6 قد سجلت افضل النتائج فيما يتعلق بالصفات الانتاجية التي شملتها الدراسة الحالية مقارنة بالمعاملات T3 و T4 و T5.

يستنتج من الدراسة الحالية بأن اضافة مستخلص عرق السوس خصوصاً بمستوى 450 ملغم/كغم للعلف الملوث بسموم الافلاتوكسين يمكن ان يحد من التأثيرات الضارة لهذه السموم في الأداء الإنتاجي لفروج اللحم.

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Introduction

The aflatoxin-producing fungi have been found in large variety of commodities. Condition favouring their growth and toxin production are : high moisture content, high temperature, insect damage, and the physical condition of the grain, weathering, mechanical handling and presence of cracked grains (13) .Reddy et al ., (15) reported that infestation of feeds with aflatoxins causes increased mortality, decreased growth rate and poor feed conversion in broiler chicken .However, aflatoxin was demonstrated to produce in broiler chicken a steatorrhea accompanied by a decrease in digestive enzymes elaborated by pancreas(14). Giambrone et al., (7) indicated that aflatoxin treatment resulted in increase in the susceptibility of broiler chickens to salmonellosis, aspergillosis, coccidiosis, and Marek's disease. The immuno depressive action in aflatoxins was primarily on the cell-mediated immune system (7).

Aflatoxin is the common name for a group of structurally related compounds (aflatoxin B1, B2,G1, and G2) produced by fungi of the *flavus – parasiticus* group of the genus *Aspergillus* .This mycotoxin is potentially a threat to poultry health and production through contamination of poultry feeds (8). Al-Daraji (2) reported that experimentally induced aflatoxicosis resulted in significant deterioration in erythrocytes, leucocytes, thrombocytes, haemoglobin concentration, heterophil/ lymphocyte ratio, hematocrit and plasma uric acid, glucose, cholesterol, protein, calcium, phosphorus, GOT activity and alkaline phosphatase activity.

Licorice exerts numerous beneficial effects on the body, making licorice a valuable herb for treating a host of aliments. It can help reduce inflammation. It seems to prevent the breakdown of adrenal hormones such as cortisol (the body's primary stress – fighting adrenal hormone), making these hormones more available to the body and helps the body cope with stress (20). Licorice also appears to enhance immunity by boosting levels of interferon, a key immune system chemical that fights off attacking viruses (6) . However, licorice is also known to exhibit many pharmacological actions, including estrogenic activity, anti – inflammation, anti allergic, antibacterial, antiviral, antihepatotoxic, fungicide, anticancer and anti – Trichomonas (12).

The present study was undertaken as a trial to suppress the effect of aflatoxicosis on productive performance of broiler chickens by using different levels of licorice extract. Licorice extract was supplemented at levels of 150, 300, and 450 mg/kg to the diet of birds which was previously contaminated with aflatoxin.

Materials and Methods

This study was conducted at the Animal Production farm/State Board for Agricultural Research over the period from 21/3/2002 to 27/4/2002 .A total of 900 Fawbro broiler, three weeks of age were used. Birds were fed starter diet during the third week of age (beginning date of experiment; 22.7% crude protein and 2867.4 kcal/kg of diet) and finisher diet (20.6% crude protein and 2922 kcal/kg of diet) until the marketing age (49 days of age). Chicks were randomly divided into 6 treated groups of 3 replicate per group, each replicate constitutes 50 chicks (150 chicks per treatment group).

Birds in the first treatment fed a commercial broiler ration and used as a control group (T1) .The second treatment (T2) was fed a diet contaminated with aflatoxin, while birds in the third treatment (T3) were fed a diet contaminated with aflatoxin and treated with mold killer (Choong ang Biotech company, Korea). However, birds in fourth, fifth, and sixth treatments were fed a diet contaminated with aflatoxin and supplemented with licorice extract. Licorice extract was supplemented to the diet of birds throughout the total period of experiment at levels of 150 mg/kg (T4), 300 mg/kg (T5) and 450 mg/kg of diet (T6).

Aflatoxin used in the present study was aflatoxin B1 which obtained from the Department of Plant Protection, College of Agriculture, University of Baghdad. Aflatoxin was prepared and incorporated into basal diet by method previously reported (17). Aflatoxin was produced by growing *Aspergillus flavus* on rice. The moldy rice was dried and ground to a fine powder and analyzed spectrophotometrically for its total aflatoxin content by the method of Nabney and Nesbitt (10). The moldy rice then added to the yellow corn that involved in the basal diet. The final level of aflatoxin introduced to the birds was determined to be equal to 2 mg aflatoxin/kg of diet.

Productive characteristics measured in this study included: body weight, weight gain, feed consumption, feed conversion ratio, and mortality. However, Productive Index and Economic Figure were determined according to Naji and Hana (11). At the end of experiment, 18 birds per each treatment (6 birds per each replicate) were sacrificed to determine dressing percentage with or without

viscera, and weights of visceral organs, *viz.* liver, heart, gizzard, spleen, in addition to abdominal fat .

Significance of data was determined at the 5% level of probability by analysis of variance (ANOVA) using the Statistical Analysis of System (16). Significance of the differences between treatment means was determined by Duncan's multiple range test (16).

Results and Discussion

Dietary aflatoxin (T2) significantly ($P<0.05$) depressed body weight, weight gain, feed consumption, feed conversion, Productive Index, Economic Figure, and liveability starting from the fourth week of age through the seventh week of age in comparison with control group (T1; Figures 1, 2, 3, 4 and Table 1). When mold killer (T3) or licorice extract (T4, T5 and T6) were added to the diet containing aflatoxin, they significantly ($p<0.05$) increased these traits. However, T4, T5, and T6 surpasses T3 treatment as regards live body weight and weight gain (Figures 1 and 2). There were no significant differences between licorice treatments (T4, T5, and T6) and T3 throughout the experimental period with relation to feed conversion efficiency and mortality (Figure 4 and Table 1). Furthermore, there were no significant differences between licorice treatments and T3 treatment during 4th and 5th weeks of age, between T3, T4 and T5 treatments during the 6th week of age, and between T3 and T4 treatments during the 7th week of age in regard to feed consumption (Figure 3).

The addition of 450 mg/kg licorice extract to the diet containing aflatoxin restore the mean of live body weight, feed consumption, Productive Index and Economic Figure to the control values (Figures 1, 2, 3 and Table 1).

The results of this investigation clearly demonstrate that aflatoxicosis in broiler chickens can be influenced by supplementation the licorice extract to the contaminated diet .Increasing the licorice content of the diet to 450 mg/kg essentially negated the effects of aflatoxin. An obvious explanation to the protective effects of licorice is that licorice shows some anti–infective properties .In laboratory and animal studies, it has stopped or slowed down the growth of certain bacteria, fungi, and parasites. Several animal studies have also revealed a possibly strong antiviral and fungicide effects for true licorice (5) .In these studies, true licorice component that belong to the isoflavonoid class of chemicals, appear to have several anti – infective effects that include interference with oxygen utilization by infective – organisms. Additionally, true licorice may have some ability to improve functioning of the immune system (1, 18). Newall et al. (12) reported that medicinal use of licorice in both Western and Eastern cultures dates back several thousands years. Licorice is know to exhibit many pharmacological actions, including anti–inflammatory (cortisol–like), antiviral, antibacterial, antifungal, anti-Trichomonas, antihepatot-oxic and anti allergic activities. The plant reinforce the body's ability to withstand attack from virtually any kind of pathogen. However, if one is looking for a broad-spectrum tonic to protect, maintain health, and heal injuries, there is no herb better than licorice root. Utsunomiya et al. (20) indicated that modern research on licorice reports many effects which are adrenal enhancing, analgesic, anti inflammatory, antioxidant, anti tumor, antiviral, fungicide, immune protecting, liver protecting and liver detoxifying. However, by functioning as anti – fungal agent, this herb destroys or prevent the growth of fungi.

Inclusion the aflatoxin in the diet (T2) resulted in a significant reduction in both dressing percentage with or without viscera compared with control group (T1 ; Table 1) . However, the supplementation of mold killer (T3) and licorice extract (T4, T5, T6) to the aflatoxin – contaminated diet significantly improved these two traits in comparison with T2 treatment . T6 treatment surpasses other treatments in relation to dressing percentage with or without viscera and it restore the means of these two traits to the control values.

The effects of different treatments on the relative weight of certain organs are presented in Table 1. With incorporation of aflatoxin into the diet (T2), visceral organs such as liver, heart, gizzard and spleen significantly ($p < 0.05$) increased in comparison with control group (T1) . However, administration of graded levels of licorice extract (T4, T5, T6) or mold killer (T3) resulted in significant reduction in the relative weight of these organs compared with T2 treatment. Furthermore, there was a trend that T6 recorded the lowest ($p< 0.05$) means regarding the relative weights of liver, heart, gizzard and spleen comparing with other treatments (T3, T4 and T5).

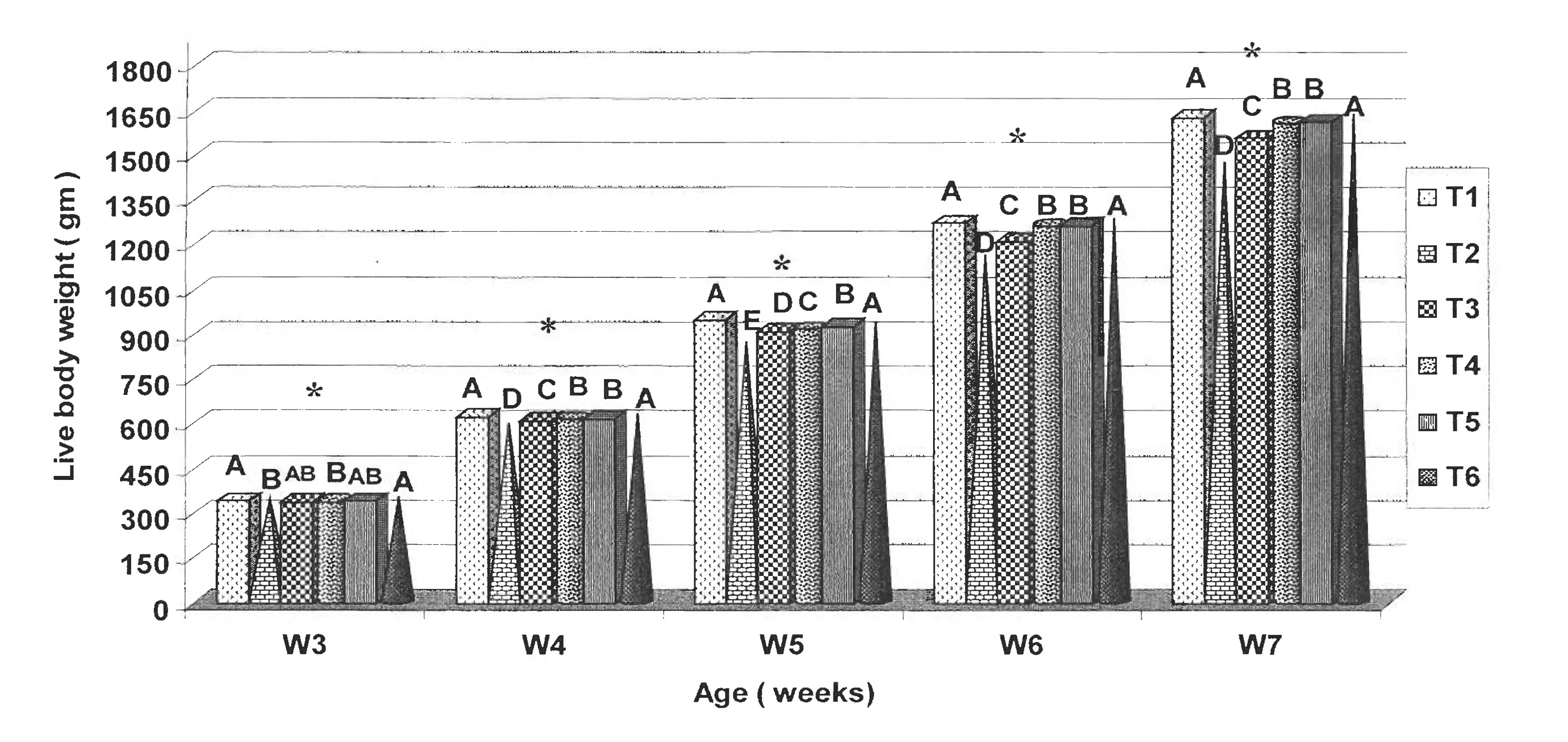


Figure 1.The effect of different levels of licorice extract on mean body weight of broiler fed a diet contaminated with aflatoxin .

T1= Birds fed a basal diet , T2= Birds fed diet contaminated with aflatoxin , T3=Birds fed diet contaminated with aflatoxin and treated with mold killer , T4= Birds fed diet contaminated with aflatoxin and supplemented with licorice extract at level of 150 mg / kg , T5= Birds fed diet contaminated with aflatoxin and supplemented with licorice extract at level of 300 mg / kg , T6= Birds fed diet contaminated with aflatoxin and supplemented with licorice extract at level of 450 mg / kg.

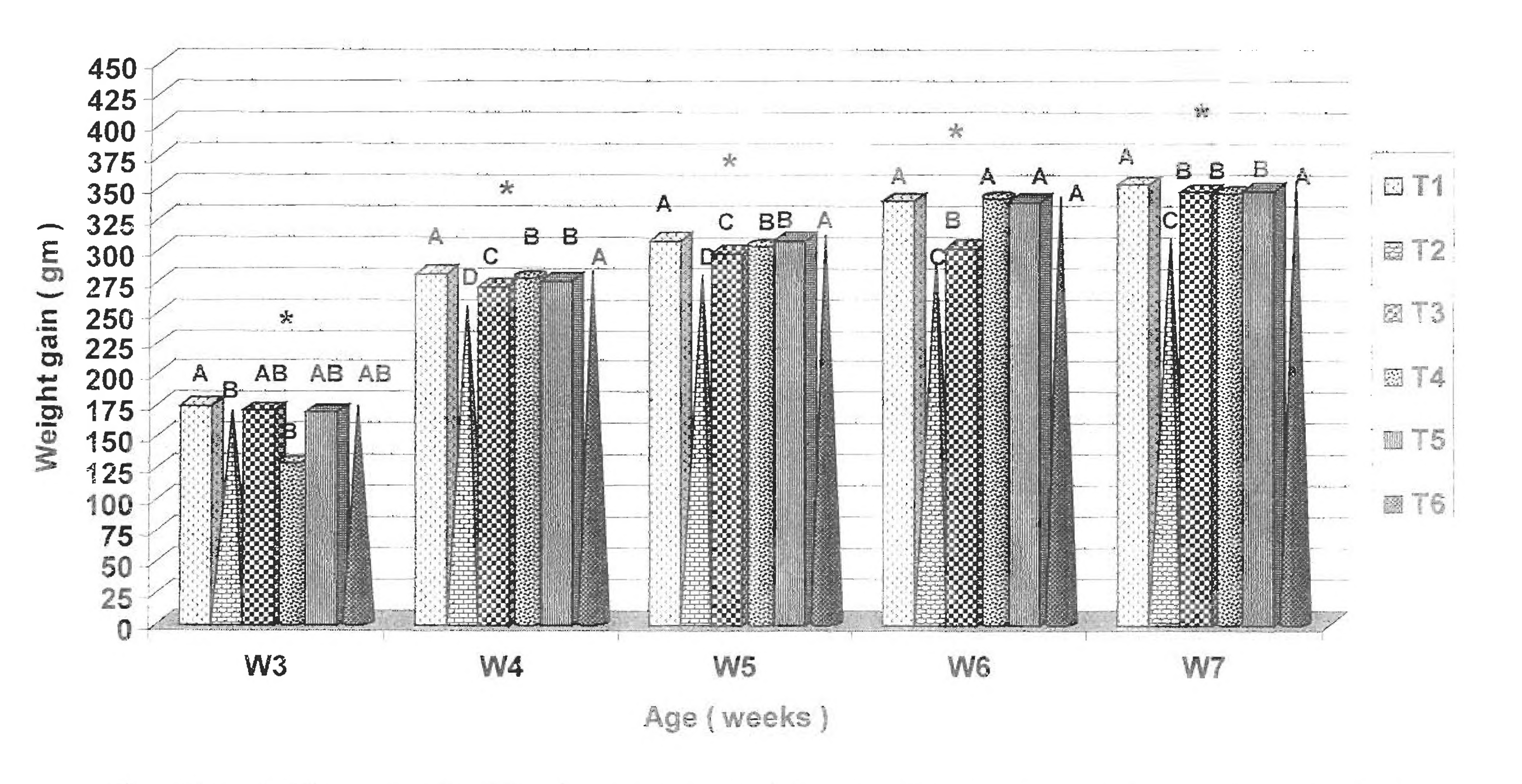


Figure 2.The effect of different levels of licorice extract on weight gain of broiler fed a diet contaminated with aflatoxin

T1= Birds fed a basal diet , T2= Birds fed diet contaminated with aflatoxin , T3=Birds fed diet contaminated with aflatoxin and treated with mold killer , T4= Birds fed diet contaminated with aflatoxin and supplemented with licorice extract at level of 150 mg / kg , T5= Birds fed diet contaminated with aflatoxin and supplemented with licorice extract at level of 300 mg / kg , T6= Birds fed diet contaminated with aflatoxin and supplemented with licorice extract at level of 450 mg/kg.

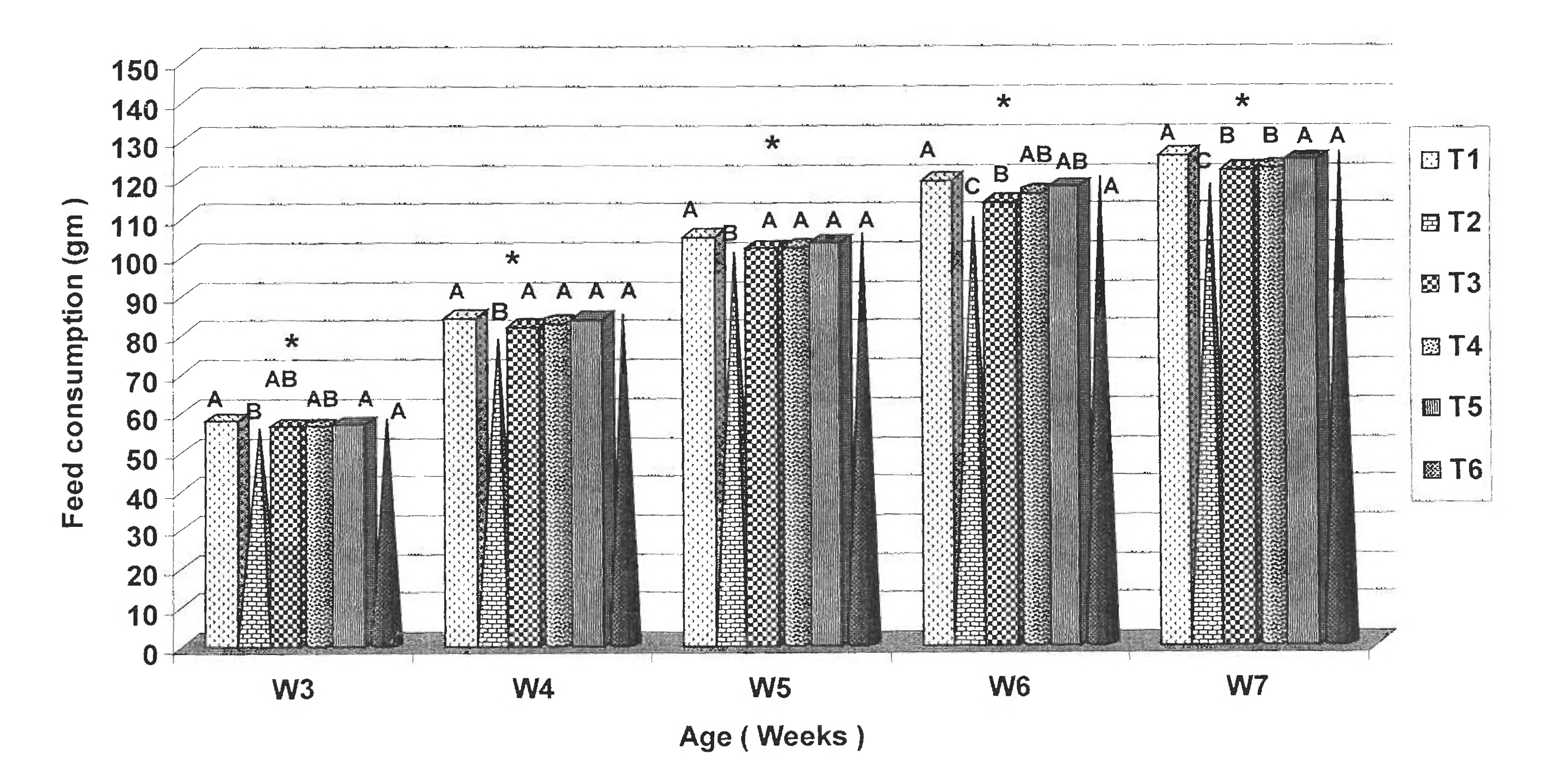


Figure 3.The effect of different levels of licorice extract on feed consumption of broiler fed a diet contaminated with aflatoxin .

T1= Birds fed a basal diet , T2= Birds fed diet contaminated with aflatoxin , T3=Birds fed diet contaminated with aflatoxin and treated with mold killer , T4= Birds fed diet contaminated with aflatoxin and supplemented with licorice extract at level of 150 mg / kg , T5= Birds fed diet contaminated with aflatoxin and supplemented with licorice extract at level of 300 mg / kg , T6= Birds fed diet contaminated with aflatoxin and supplemented with licorice extract at level of 450 mg/ kg.

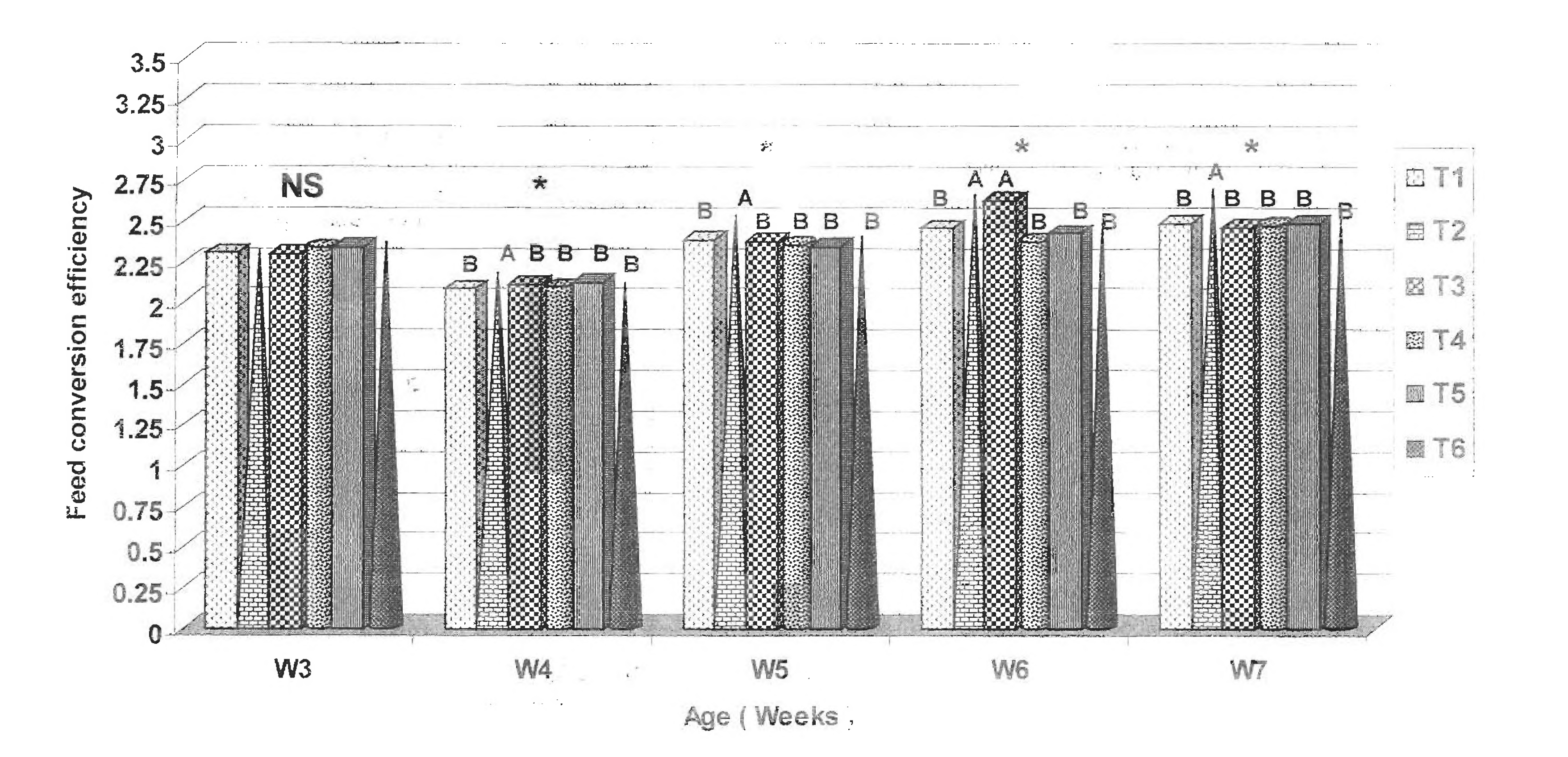


Figure 4.The effect of different levels of licorice extract on feed conversion efficiency of broiler fed a diet contaminated with aflatoxin

T1= Birds fed a basal diet , T2= Birds fed diet contaminated with aflatoxin , T3=Birds fed diet contaminated with aflatoxin and treated with mold killer , T4= Birds fed diet contaminated with aflatoxin and supplemented with licorice extract at level of 150 mg / kg , T5= Birds fed diet contaminated with aflatoxin and supplemented with licorice extract at level of 300 mg / kg , T6= Birds fed diet contaminated with aflatoxin and supplemented with licorice extract at level of 450 mg / kg.

Table 1. The effect of different levels of licorice extract on certain productive performance and organ weights of broiler fed a diet contaminated with aflatoxin

Traits \ Treatments	T1	T2	T3	T4	T5	T6
Productive Index	A 195.28 ± 12.21	D 161.60 ± 15.69	C 180.55 ± 14.53	BC 183.14 ± 13.9	B 185.02 ± 14.03	AB 190.18 ± 12.71
Economic Figure	A 196.04 ± 13.47	D 162.14 ± 17.15	C 181.08 ± 15.32	BC 183.63 ± 16.01	B 185.49 ± 15.44	AB 191.01 ± 14.8
Total mortality (%)	C 1.39 ±0.06	A 2.69 ± 0.11	B 1.62 ± 0.08	B 1.59 ± 0.08	B 1.48 ± 0.06	B 1.41 ± 0.05
Dressing percentage (with viscera)	A 72.81 ± 4.69	C 71.93 ± 7.33	B 72.55 ± 6.28	B 72.53 ± 6.19	B 72.74 ± 5.63	A 74.80 ± 4.82
Dressing percentage (without viscera)	A 65.14 ± 3.95	C 63.92 ± 5.64	B 64.72 ± 4.63	B 64.77 ± 4.49	B 64.97 ± 5.02	A 65.1 ± 4.11
Abdominal fat (%)	C 1.04 ± 0.09	A 1.11 ± 0.01	AB 1.09 ± 0.01	C 1.05 ± 0.01	B 1.08 ± 0.08	C 1.05 ± 0.01
Liver weight (%)	D 3.01 ± 0.08	A 3.89 ± 0.16	B 3.17 ± 0.07	B 3.16 ± 0.09	B 3.15 ± 0.09	C 3.05 ± 0.07
Heart weight (%)	D 0.51 ± 0.05	A 0.68 ± 0.01	B 0.60 ± 0.08	B 0.60 ± 0.09	B 0.58 ± 0.08	C 0.54 ± 0.06
Spleen weight (%)	D 0.10 ± 0.003	A 0.28 ± 0.009	B 0.17 ± 0.005	B 0.16 ± 0.005	B 0.15 ± 0.004	C 0.12 ± 0.03
Gizzard weight (%)	D 2.60 ± 0.01	A 3.03 ± 0.03	B 2.84 ± 0.02	B 2.79 ± 0.02	B 2.81 ± 0.02	C 2.66 ± 0.02

***T1= Birds fed a basal diet, T2= Birds fed diet contaminated with aflatoxin, T3=Birds fed diet contaminated with aflatoxin and treated with mold killer, T4=Birds fed diet contaminated with aflatoxin and supplemented with licorice extract at level of 150mg/kg, T5= Birds fed diet contaminated with aflatoxin and supplemented with licorice extract at level of 300 mg/kg, T6= Birds fed diet contaminated with aflatoxin and supplemented with licorice extract at level of 450 mg/kg.**

****Values in a row with different superscripts differ significantly ($p < 0.05$) .**

Liver seemed to be the target organ in aflatoxicosis. Hepatic hypertrophy following toxin administration appeared to be caused primarily by increased fat content in the organ (15). Mashaly et al.(9) reported that aflatoxin feeding at 50 μg/kg diet resulted in a significant decline in body and liver weights, rate of liver protein and RNA synthesis, and muscle RNA synthesis .Brown and Abrams (4) observed that a severe decline in plasma proteins on feeding aflatoxin B1 to chickens and ducklings was due to the suppression of liver protein synthesis as a consequence of mitochondrial injury and the lowered rate of ATP synthesis . Huff et al .(8) also noted that aflatoxin treatment significantly ($p < 0.05$) decreased body weight and weight gain ; increased the relative weight of the spleen, liver, proventriculus, gizzard, heart, and kidney ; and induced hepatic hyperlipemia . Reddy et al . (15) found that with the increase of the level of aflatoxin, liver kidney, spleen, gizzard and pancreas showed an increase in weight with respective threshold doses of 0.50, 0.75, 1.50 and 4.0 ppm, while bursa of fabricius regressed at 1.25 ppm . Smith and Hamilton (19) demonstrated that graded doses of aflatoxin (1.25, 2.5, 5.0 and 10.0 ppm) incorporated into the feed of broiler chickens resulted in a decreased growth rate, an enlarged liver, spleen, and pancreas and a regressed bursa of fabricius. However, analysis of the liver showed that lipids accounted for 60 % of the dry weight increase (19).

The finding that licorice extract can suppress the detrimental effects of aflatoxin on liver may be explained by that active component in licorice root are also used to help prevent and treat chronic hepatitis (liver inflammation). In one study of Japanese patients with hepatititis C those received intravenous treatment with glycyrrhizin for an average of 10 years were significantly less likely to develop liver cancer and cirrhosis (progressive liver failure ; 21). In a second study of 57 patients with hepatitis C, glycyrrhizin (in dose ranging from 80 to 240 mg/day) significantly improved liver function after only one month. These effects diminished after glycyrrhizin treatment discontinued, however (3). Fujioka et al. (6) reported that licorice both protects the liver and promotes healing of this vital organ . The herb's anti – inflammatory properties help calm hepatitis-associated liver inflammation. Licorice also fights the virus and toxins commonly responsible for hepatitis, and supplies valuable antioxidant compounds that help maintain the overall health of liver and certain vital organs. However, glycyrrhizin may protect liver and other vital organs such as heart, spleen, and kidney from being damaged by oxidants. Too many oxidants can harm healthy cells and cause inflammation. Licorice root nutritionally supports the respiratory and gastrointestinal systems, liver, heart and spleen (22).

It was concluded from this study that dietary licorice extract especially at level of 450 mg/kg of diet can influence the severity of aflatoxicosis in broiler chickens and may be helpful in the control and prevention of this economically important disease .

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